

Amendments to the Claims:

This listing of claims will replace all prior versions, and listings, of claims in the application.

Listing of Claims:

1. (currently amended) A method of directly detecting a donor-product produced in of a group transfer reaction, the method comprising:
 - a) reacting ~~an activated form of~~ a donor molecule with an acceptor in the presence of a catalytically active enzyme;
 - b) forming the donor-product and an acceptor-X;
 - c) contacting the donor-product with a first complex comprising a macromolecule, that specifically recognizes the donor-product, and ~~comprising~~ a detectable tag capable of producing an observable;
 - d) competitively displacing the detectable tag of the first complex by the donor-product to generate a second complex and a displaced detectable tag to directly detect the donor-product in the group transfer reaction without first removing or altering the residual donor molecule; and
 - e) detecting a change in the observable produced by the detectable tag in the first complex and the displaced detectable tag.
2. (original) The method of Claim 1, further comprising,
 - f) quantifying the observable of step (e).

3. (currently amended) The method of Claim 1, wherein,
- a) ~~the activated form of the donor molecule~~ comprises a donor-X,
wherein the X is a covalent adduct, a phosphate;
 - b) the acceptor comprises a substrate for the catalytically active enzyme,
wherein the substrate is ~~selected from the group consisting of a polypeptide, a protein, a nucleic acid, a lipid, a carbohydrate and a small molecule substrate;~~
 - c) the donor-product comprises a nucleotide ~~or a non-nucleotide, wherein the non-nucleotide is a metabolic intermediate selected from the group consisting of s-adenosylhomocysteine, nicotinamide or coenzyme A;~~
 - d) the acceptor-X comprises a reaction product in which X is a covalent adduct, ; ~~wherein the covalent adduct is selected from the group consisting of a phosphate, a sulfate, a carbohydrate, a naturally occurring amino acid, a synthetically derived amino acid, ADP-ribose, a nucleotide, a methyl, an acetyl, and a glutathione moiety;~~ and wherein the covalent adduct is optionally capable of altering either the function, the stability, or both the function and the stability of the acceptor;
 - e) the first complex comprises a macromolecule and a detectable tag; and
 - f) the second complex comprises the macromolecule wherein the detectable tag is competitively displaced by the donor-product resulting in the production of an observable.
4. (currently amended) The method of Claim 3, wherein the macromolecule is ~~selected from the group consisting of an antibody, a polypeptide, a protein, a nucleic acid molecule, and an inactivated enzyme that is capable of contacting the donor-product with high affinity.~~
5. (currently amended) The method of Claim 4, wherein the antibody is a monoclonal antibody, ~~a polyclonal antibody, or a recombinant antibody.~~

6. (currently amended) The method of Claim 4, wherein the antibody is specific for the donor-product, and wherein the level of antibody cross-reacting with the donor-X is less than the level of specificity that the antibody exhibits towards the donor-product.

7. (currently amended) The method of Claim 1, wherein the detectable tag is a tracer, wherein the tracer is a fluorescent ~~or a chemiluminescent~~ molecule conjugated to a nucleotide ~~or a non-nucleotide~~.

8. (original) The method of Claim 1, further comprising detecting a catalytic activity, wherein the catalytic activity generates the donor-product in the group transfer reaction.

9. (currently amended) The method of Claim 8, wherein the catalytic activity comprises ~~a chemical catalytic activity, an enzymatic activity, or a combination thereof;~~ wherein the enzymatic activity comprises ~~a sulfotransferase, a kinase, a UDP-glucuronosyltransferase, a methyl transferase, a acetyl transferase, a glutathione transferase, and a ADP-ribosyltransferase.~~

10 (original) The method of Claim 1, wherein the method is an immunoassay.

11. (currently amended) The method of Claim 10, wherein the immunoassay is ~~selected from the group consisting of fluorescence polarization immunoassay (FPIA); fluorescence resonance energy transfer (FRET); enzyme linked immunosorbant assay (ELISA); chemiluminescence immunoassay.~~

12. (original) The method of Claim 1, wherein the method is used for screening a chemical library to identify a molecule which is capable of activating or inhibiting a group transfer reaction enzyme.

13. (original) The method of Claim 12, wherein the molecule is capable of altering either the function, the stability, or both the function and the stability of the acceptor.

14. (original) The method of Claim 12, wherein the molecule is capable of exhibiting a therapeutic effect.

15. (original) The method of Claim 12, wherein the library is screened using a high-throughput screening technique comprising a multiwell plate, a microarray or a microfluidic system.

16. (withdrawn) An antibody produced against a donor product of a group transfer reaction, wherein the antibody comprises the ability to preferentially distinguish between a donor-product and a donor in the presence of a high donor concentration.

17. (withdrawn) The antibody of Claim 16, wherein the donor-product is selected from the group consisting of a nucleotide or a non-nucleotide.

18. (withdrawn) The antibody of Claim 16, wherein the antibody is specific for a phosphate portion of a nucleotide, and wherein the antibody has the ability to distinguish between a 5'-phosphate, a 5' -phosphosulfate, a 5'-diphosphate and a 5' -triphosphate.

19. (currently amended) A ~~homogeneous competitive binding assay for method~~ of directly detecting a donor-product produced in of a group transfer reaction without first removing or altering a residual donor molecule, the assay method comprising the steps of:

- a) combining the donor-product produced in a group transfer reaction with a tracer and a macromolecule to provide a reaction mixture, the macromolecule being specific for the donor-product, the tracer comprising the donor-product conjugated to a fluorophore, ~~the tracer and capable of binding being able to bind~~ to the macromolecule to produce a detectable change in fluorescence polarization;
- b) measuring the fluorescence polarization of the mixture to obtain a measured fluorescence polarization; and
- c) comparing the measured fluorescence polarization with a characterized fluorescence polarization value, ~~the characterized fluorescence polarization value~~ corresponding to a known donor-product concentration to directly detect the donor-product produced in the group transfer reaction.

20. (currently amended) The assay of Claim 19, wherein the group transfer reaction is catalyzed by an enzyme.

21. (currently amended) The assay of Claim 19, wherein the enzyme is ~~selected from the group consisting of a kinase, a sulfotransferase, a methyltransferase a UDP-glucuronosyltransferase, a acetyl transferase, a glutathione transferase, and a ADP-ribosyltransferase.~~

22. (currently amended) The assay of Claim 19, wherein the donor-product is ~~selected from the group consisting of phosphoadenosine phosphosulfate (PAP), adenosine diphosphate (ADP), uridine diphosphate (UDP), s-adenosylhomocysteine, nicotinamide, and Coenzyme A.~~

23. (currently amended) The assay of Claim 19, wherein the fluorophore is ~~selected from the group~~ fluorescein, preferably Alexa Fluor® dyes rhodamine, Texas red and derivatives thereof.

24. (original) A method of using the assay of Claim 19 to screen a chemical library to identify a molecule which is capable of inhibiting or activating a group transfer reaction enzyme.

25. (withdrawn) An assay kit for characterizing a donor-product from a group transfer reaction, the assay kit comprising:

a macromolecule and a tracer, each in an amount suitable for at least one homogeneous fluorescence polarization assay for donor-product, wherein the macromolecule is an antibody or an inactivated enzyme; and wherein the macromolecule and the tracer may be separate or together in the container.

26. (withdrawn) The assay kit of Claim 25, further comprising packaging, and instructions for using the antibody and the tracer in the homogeneous fluorescence polarization assay, the antibody being specific for donor-product, the tracer comprising donor-product conjugated to a fluorophore, the tracer being able to bind to the antibody to produce a detectable change in fluorescence polarization.

27. (withdrawn) The assay kit of Claim 26 wherein the fluorophore is selected from the group consisting of fluorescein, rhodamine, Texas red and derivatives thereof.

28. (new) A method of directly detecting a donor-product produced in a group transfer reaction, the method comprising:

- a) reacting a donor molecule, an adenosine triphosphate (ATP), with an acceptor, a polypeptide, in the presence of a catalytically active enzyme, a kinase;
- b) forming the donor-product, an adenosine diphosphate (ADP) and an acceptor-X, a phosphorylated polypeptide;
- c) contacting the ADP with a first complex comprising a macromolecule, an antibody, that specifically recognizes the ADP and a detectable tag, a tracer, capable of producing an observable;
- d) competitively displacing the detectable tag of the first complex by the donor-product, ADP, to generate a second complex, ADP-antibody complex and a displaced detectable tag, a tracer, to directly detect the donor-product in the group transfer reaction without first removing or altering the residual donor molecule; and
- e) detecting a change in the observable produced by the tracer in the first complex bound to the antibody and the tracer.

29. (new) A method of directly detecting a donor-product produced in a group transfer reaction without first removing or altering a residual donor molecule, adenosine triphosphate (ATP), the method comprising the steps of:

- a) combining the donor-product, an adenosine diphosphate (ADP), produced in a group transfer reaction, with a tracer and a macromolecule, an antibody to provide a reaction mixture, the antibody being specific for the ADP, the tracer comprising the ADP conjugated to a fluorophore and capable of binding to the antibody to produce a detectable change in fluorescence polarization;
- b) measuring the fluorescence polarization of the reaction mixture to obtain a measured fluorescence polarization; and
- c) comparing the measured fluorescence polarization with a characterized fluorescence polarization value corresponding to a known ADP concentration to directly detect the donor-product, ADP produced in the group transfer reaction, a kinase reaction.